

# A Review of the Literature on the Etiology of Hodgkin's Disease

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## SUMMARY

*Extensive research to trace the cause of Hodgkin's disease to a bacterial or protozoan agent has proven fruitless.*

*Although a viral cause for Hodgkin's disease has been previously suggested, early explorations along that line have not been confirmed. With the development of newer techniques for the study of viral characteristics certain apparently significant factors in Hodgkin's disease have been encountered. Most promising has been the consistent demonstration that Seitz-filtered, sterile Hodgkin's disease lymph node extract can be passed serially in fertile chicken eggs and that the amniotic fluid from these eggs possesses the capacity to interfere with the growth of influenza virus in eggs.*

IT is possible to review the literature on the cause of Hodgkin's disease even though no cause has ever been generally accepted as being valid. This paradox exists because many so-called "causes" of Hodgkin's disease have been announced or strongly implied in the literature. Each of these has been followed by a rash of clinical trials of different substances supposed to neutralize or control the so-called etiologic agent, and finally further experimental and therapeutic studies (Tables 1 and 2) have given general discredit to the whole announcement.

This cycle of events has occurred so repeatedly in the case of Hodgkin's disease that a review of this phenomenon is interesting. In addition there is possibility that a small thread of truth and reason may be tangled in the maze of generally unacceptable etiologic announcements.

In general the clinical impression in Hodgkin's disease is one of a toxic or septic process coincident with recurrently enlarging lymph node tumor masses. That this frequent association of the disease with fever and toxicity has had its effect on etiologic thinking is seen in the fact that, when a specific cause has been postulated, it has almost invariably been on the basis of a supposed microbiologic organism. Hodgkin himself confused the disease with tuberculosis, and Sternberg<sup>24</sup> for a long time

felt that it was a peculiar form of tuberculosis. Later he agreed with Reed<sup>20</sup> that it was a separate entity from tuberculosis, although often associated with it, and probably itself essentially inflammatory in origin.

The possibility that Hodgkin's disease was related to tuberculosis remained in the minds of many workers, since it is known that in from 10 to 20 per cent of cases there is evidence of either inactive or active tuberculosis at autopsy. Furthermore, the negative tuberculin reaction in patients with Hodgkin's disease was so frequent as compared to negative reaction in many other diseases (Steiner<sup>23</sup>) that students could not avoid thinking of Hodgkin's disease as being perhaps an anergic phase of tuberculosis. Fraenkel and Much<sup>9</sup> and Ewing<sup>6</sup> were impressed with this apparent relationship and reported finding acid-fast rods and granules on occasion in tissue from patients who had Hodgkin's disease. In general, however, animal inoculations with such material did not produce the disease. L'Esperance<sup>15</sup> reawakened the tuberculosis theories when she postulated that the bacillus involved was of the avian strain. Although she reported producing tuberculosis in chickens by intravenous injection of ground lymph nodes from subjects with Hodgkin's disease, this was not a constant finding. Subsequent experimenters (Van Rooyen<sup>27</sup>) were unable to confirm these findings, and in general the entire concept was abandoned.

The general significance of the diphtheroid bacilli in Hodgkin's disease was first postulated (erroneously) by Bunting and Yates.<sup>4</sup> They cultured Hodgkin's disease lymph nodes in a search for an etiologic agent and isolated a diphtheroid bacillus on several occasions. They injected this organism subcutaneously into monkeys and regional lymph node disease resulted. Although the animals were toxic and the nodes frequently necrotic, the investigators nonetheless concluded that the bacillus caused a reaction "unquestionably related to human Hodgkin's disease." Subsequently many other investigators, including Fox,<sup>8</sup> demonstrated that such diphtheroids could be isolated from many types of human lymph node tissues. It was soon recognized that the reaction produced in monkeys was simply inflammatory lymphadenopathy.

Parsons and Poston<sup>19</sup> called attention to the simultaneous existence of Hodgkin's disease and brucella infections in their material. They isolated brucella from the lymph nodes in 14 consecutive cases of Hodgkin's disease (Wise and Poston<sup>28</sup>) and stated that the pathologic change in the glandular type of

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TABLE 1.—*Supposed "Etiologic Organisms" in Hodgkin's Disease*

Organism	Investigators and Date	Basis of Postulation	Evidence Against
Human tubercle bacillus	Sternberg (1898)	Similar in appearance, occasional association.	Animal inoculations negative.
Avian tubercle bacillus	L'Esperance (1931)	Tubercle bacillus in some chickens after intravenous injection of ground Hodgkin's disease nodes.	Van Rooyen (1933) inoculated chickens, with negative results.
Diphtheroids	Bunting & Yates (1914)	Isolated from Hodgkin's disease, lymph nodes; injected in monkeys, caused Hodgkin's disease.	Often found in any lymph nodes (Stewart, 1932). Did not cause Hodgkin's disease in monkeys (Fox 1915).
Ameba	Kofoed	Found ameba in lymph nodes in Hodgkin's disease.	Never confirmed. He mistook macrophages for ameba.
Brucella	Parsons & Poston (1939)	Frequently finding Brucella in lymph nodes in Hodgkin's disease.	Findings not confirmed in other part of U.S.A. (Hoster, Doan et al. 1944). It is an endemic disease in South.
Virus	Gordon (1932) Grand (1942) Bostick (1947) Bostick (1949)	Inclusion bodies. Inclusion bodies. Lethal factor for eggs. Virus interference phenomenon.	Never started Koch's postulates. No epidemiological evidence. Human inoculation unsuccessful.
Torula histoplasmosis, bacilli and cocci. Monilia, gas bacilli, yeasts	Various authors	Sometimes associated with Hodgkin's disease.	Not proposed as "cause"—only as a "trigger mechanism."

brucellosis could not be distinguished microscopically from those caused by Hodgkin's disease. Even with brucella infections patients with Hodgkin's disease often showed no or minimal reaction to the organisms as judged by immunological techniques. Experiments by the other investigators designed to confirm these findings were essentially unsuccessful. Hoster, Doan and Schumacher<sup>12</sup> cultured tissue from 45 cases of Hodgkin's disease and were unable to demonstrate brucella organisms. Further investigation revealed that brucellosis is an endemic infection in the southern area, where Parsons and Poston worked.

Several investigators have been impressed by the diversity of the types of microorganisms which have been found in lymph nodes affected by Hodgkin's disease. Billings and Rosenow<sup>1</sup> concluded that in the early phases of the disease bacilli predominate in the nodes, whereas in the later stages the cocci gain ascendancy. Twort<sup>26</sup> found many types of bacteria in cultures of lymph node tissue from subjects with Hodgkin's disease. Jackson and Parker<sup>14</sup> even found in many affected nodes a Gram-positive anaerobic gas bacillus, but the organism was also observed in some control lymph nodes. Yeasts have been isolated from about 55 per cent of lymph nodes, both in controls and those affected by Hodgkin's disease. Torulosis has been associated with Hodgkin's disease (Fitchett and Weidman<sup>7</sup>). Miller, Keddie, Johnstone and Bostick<sup>18</sup> noted the pronounced histologic similarity and coincidence of Hodgkin's disease and histoplasma capsulatum infections.

This multiplicity of organisms that have been encountered in Hodgkin's disease has induced several investigators to suggest, as did Desjardins,<sup>5</sup> that "the factor immediately responsible for lymph-

oblastomatous hyperplasia of the lymphoid structures is chronic infection of any kind." Fitchett and Weidman<sup>7</sup> considered this to be a possibility. It is known that occasionally Hodgkin's disease develops in a patient after many years of lymphadenopathy. Often in such cases there is history of recurrent transient swelling of lymph nodes, apparently bacterial in origin, and hypertrophied tonsils. Whether or not such a "trigger" action of chronic non-specific bacterial lymphadenitis exists in certain predisposed patients is not known, although it is neither impossible nor unreasonable.

Bacterial and animal inoculation studies (Table 2) of the Hodgkin's disease process did little to clarify the problem and search for a viral agent developed. Gordon<sup>10</sup> and his associates made the first systematic investigation in this regard, although earlier investigators (Twort<sup>26</sup> and McJunkin<sup>16</sup>) and many later ones concluded that viral infection would offer a logical explanation of the whole process. These conclusions were based upon theoretical considerations reached in the light of known clinical, pathological and experimental characteristics of Hodgkin's disease, which at times possess a resemblance to characteristics of certain known viral tumors of animals.

Gordon's investigation of Hodgkin's disease consisted of a broad study of the effect of the inoculation of various animals—including monkeys, mice, guinea pigs and rabbits—with ground tissue from subjects with Hodgkin's disease. Results of all the procedures were essentially negative except for the observation that, in rabbits, intracerebral inoculations resulted in the development of an encephalitis and convulsive syndrome which developed in from two to six days after incubation. This reaction occurred in from 60 to 75 per cent of cases in which

TABLE 2.—*Hodgkin's Disease Treatment with Biological Preparations*

Authors	Material	Results
Billings & Rosenow (1913)	Diphtheroid vaccine.	Unsuccessful.
Moore (1916)	Diphtheroid vaccine made in horses.	Not successful.
Coley (1928)	Prodigious and streptococcus toxins.	Poor.
Utz & Keatings (1932)	Chickens immunized with Hodgkin's disease serum.	Authors: "Encouraging." Others: "Negative."
Gordon (1932)	"Sensitized vaccine" against "virus inclusion bodies." Rabbits used.	Unsatisfactory.
Parsons & Poston (1939)	Brucella vaccine to build up titre in patient.	Not successful.

TABLE 3.—*Animals Inoculated with Lymph Node Mash Derived from Subjects with Hodgkin's Disease*

Animal	Author	Means of Inoculation*	Duration	Results
Chickens	L'Esperance (1931)	I.V.	1 to 10 months	Apparent tubercle bacillus developed in some.
Chickens, guinea pigs, rabbits, dogs, mice	Steiner (1934)	I.V., I.P., I.C., S.C.	.....	Negative.
Guinea pigs	Forbes & Gunther (1941)	I.P.	Variable	No specific lesions.
Guinea pigs, mice, rabbits, rats	Twort (1930)	I.P., Brain, S.C., Nasal, Oral	Months	Negative, except rare guinea pig skin reaction.
Guinea pigs, rabbits	McGrath (1933)	S.C., I.V., I.C., I.P.	.....	Negative, except rare guinea pig skin reaction.
Monkeys	Bunting & Yates (1914)	S.C. and into tonsil area	1 to 3 months	Lymph node which histologically "leaves no question as to the relation to human Hodgkin's disease."
Monkey	McGrath (1933)	S.C., I.V., I.P.	.....	Negative.
Rabbit	Gordon (1932)	All routes	Variable	Negative. "Gordon test."
Man	Tyzzar (1916)	S.C.	Weeks	No tumors resulted.

\* Key to abbreviations: I.V.—Intravenous; I.P.—Intraperitoneal; I.C.—Intracerebral; S.C.—Subcutaneous.

the inoculation material was prepared from nodes from Hodgkin's disease patients and in only about 2 per cent of cases when "control" lymph nodes were used. In the rabbits the microscopic changes were non-specific, and the reaction could not be transmitted to a second rabbit. Guinea pigs were later found to respond in a similar manner.

The encephalitogenic agent remained potent for up to two years, withstood 65° C. but not 70° C. for 30 minutes, and was filterable. These observations of Gordon were confirmed, but the same reactions were produced by inoculation with preparations made from other tissues, especially normal human bone marrow, spleen and leukocytes. Further search revealed that this encephalitogenic agent had many features in common with the proteolytic enzyme of Jockmann, a substance which was found in many normal tissues and which caused encephalitis in rabbits. Although the parallelism between the Gordon phenomenon and Jockmann proteolytic enzyme was not complete, it was sufficient to warrant the conclusion that they are essentially similar substances. A further correlation was made with eosinophilia, so frequently found in lymph nodes of patients with Hodgkin's disease. Turner, Jackson

and Parker<sup>25</sup> were able to correlate the presence of these cells with the occurrence of a positive reaction to the Gordon test, and McNaught<sup>17</sup> concluded that the Gordon test was essentially a test for the presence of eosinophils in the tissues.

Gordon also reported the presence of elementary bodies in suspensions of Hodgkin's disease lymph nodes which bore a striking morphologic resemblance to the elementary bodies of vaccinia and psittacosis. He was not able to establish any specific characteristics of the bodies which he observed and photographed. He prepared a "sensitized vaccine" to these particles, but therapeutic use of it was unsuccessful. Grand<sup>11</sup> reinvestigated the apparent elementary bodies in Hodgkin's disease by the use of tissue culture techniques. He reported that Hodgkin's disease tissue contained specific cell inclusions and that the addition of acellular Hodgkin's disease extracts to tissue culture of chorio-allantoic membranes of chicken eggs caused those specific inclusions to appear in the cultures.

Hoster and co-workers<sup>13</sup> partially repeated Grand's work but were not able to confirm his findings, and they remarked that inclusion bodies occurred in many apparently normal chicken membrane tissues.

Rottino and co-workers<sup>21</sup> also concluded that the changes were non-specific.

A thorough study of Hodgkin's disease extracts by employing the technique of fertile chicken egg passage was made by Bostick.<sup>2</sup> In early investigations it was demonstrated that sterile, Seitz-filtered amniotic fluid from Hodgkin's disease-inoculated eggs was lethal to chicken embryo in a greater percentage of instances than was control amniotic fluid. Evidence for the presence of this filterable factor was sought by many accepted procedures in virology and immunology.

Hemagglutination techniques, animal injections, cutaneous sensitivity, flocculation and precipitin tests were surveyed without significant results (Bostick<sup>3</sup>). Attention was then directed toward the capacity of this factor to interfere with the growth of known viruses in chicken eggs.

The virus interference studies encompassed a survey of many viruses. However, the most promising results were apparent initially with influenza A virus. After seven days of initial incubation, the eggs were inoculated with Hodgkin's disease amniotic fluid derived from other egg passages. After three days of further incubation, the challenging virus, influenza, was introduced. After 18 hours of incubation the amniotic fluid of all eggs was harvested and titred for the amount of influenza in them. This titre was determined by means of the hemagglutination technique of Salk,<sup>22</sup> which is based upon the fact that influenza virus is capable of hemagglutinating human erythrocytes essentially in proportion to the amount of virus present.

The Hodgkin's disease material derived from 12 separate patients interfered with the growth of influenza virus. Parallel control material showed no such capacity. This property of extracts of ground Hodgkin's disease material was retained even after 10 to 15 serial passages in fertile chicken egg amniotic sacs and after Seitz filtering. Thus, many of the characteristics of a virus are possessed by this factor, which may also be the same factor which produces the slight lethal effect on the chicken embryos. In the Hodgkin's disease-inoculated embryos no gross or microscopic evidence of pathologic change was observed. These data strongly support a postulation that there is consistently in Hodgkin's disease tissues a factor which is not present in normal or carcinomatous tissues. It is most unlikely that this factor is some incidentally isolated agent that may be present in many random lymph nodes, since it was not encountered in the control lymph node tissues. These interference phenomena offer for the first time a practical tool by which Hodgkin's disease can be extensively studied from a promising etiologic aspect.

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